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08540347 95197292

**Isolation and characterization of a family of porin proteins from Helicobacter pylori.**

Exner MM; Doig P ; Trust TJ; Hancock RE

Department of Microbiology and Immunology, University of British Columbia, Vancouver, Canada.

Infection and immunity (UNITED STATES) Apr 1995, 63 (4) p1567-72,

ISSN 0019-9567 Journal Code: GO7

Contract/Grant No.: R01AI29927-01A2, AI, NIAID

Languages: ENGLISH

Document type: JOURNAL ARTICLE

JOURNAL ANNOUNCEMENT: 9506

Subfile: INDEX MEDICUS

Two-dimensional gel electrophoresis was used to identify heat-modifiable outer membrane proteins, which were candidates for **porins**, from Helicobacter pylori membrane preparations. Four such proteins with apparent molecular masses of 48, 49, 50, and 67 kDa were isolated. The four proteins copurified together after selective detergent solubilizations followed by anion-exchange chromatography, and each protein was ultimately purified to homogeneity by gel purification. These proteins were then tested for pore-forming ability with a planar lipid bilayer model membrane system. All four proteins appeared to be present as monomers, and they formed pores with low single-channel conductances in 1.0 M KCl of 0.36, 0.36, 0.30, and 0.25 nS, respectively, for the 48-, 49-, 50-, and 67-kDa proteins which we propose to designate HopA, HopB, HopC, and HopD. N-terminal amino acid sequence analyses showed a high degree of homology among all four proteins, and it appears that these proteins constitute a family of related **porins** in H. pylori.

Tags: Support, Non-U.S. Gov't; Support, U.S. Gov't, P.H.S.

Descriptors: Helicobacter pylori--Chemistry--CH; \***Porins** --Isolation and Purification--IP; Amino Acid Sequence; Electric Conductivity; Electrophoresis, Gel, Two-Dimensional; Heat; Helicobacter pylori --Physiology--PH; Ion Channels--Chemistry--CH; Ion Channels--Isolation and Purification--IP; Molecular Sequence Data; Molecular Weight; Multigene Family; **Porins** --Chemistry--CH; Sequence Alignment; Sequence Homology, Amino Acid

CAS Registry No.: 0 (Ion Channels); 0 (Porins)

Gene Symbol: hopB; hopC; hopD; hopA

2/9/4 (Item 4 from file: 155)

08021587 95012645

**Identification of surface-exposed outer membrane antigens of Helicobacter pylori.**

**Doig P ; Trust TJ**

Department of Biochemistry and Microbiology, University of Victoria,  
British Columbia, Canada.

Infection and immunity (UNITED STATES) Oct 1994, 62 (10) p4526-33,  
ISSN 0019-9567 Journal Code: GO7

Contract/Grant No.: 1R01AI29927-01A2, AI, NIAID

Languages: ENGLISH

Document type: JOURNAL ARTICLE

JOURNAL ANNOUNCEMENT: 9501

Subfile: INDEX MEDICUS

Despite the potential significance of surface-localized antigens in the colonization by and disease processes of *Helicobacter pylori*, few such components have been unequivocally identified and/or characterized. To further investigate the surface of this bacterium, monoclonal antibodies (MAbs) to a sarcosine-insoluble outer membrane fraction prepared from *H. pylori* NCTC 11637 were raised. MAbs were selected on the basis of their surface reactivity to whole cells by enzyme-linked immunosorbent assay, immunofluorescence, and immunoelectron microscopy. By use of this selection protocol, 14 surface-reactive MAbs were chosen. These MAbs were used to identify six protein antigens (molecular masses, 80, 60, 51, 50, 48, and 31 kDa), all of which were localized within or associated with the outer membrane. Two of the MAbs recognized the core region of lipopolysaccharide (LPS). Only these two anti-LPS MAbs also recognized the flagellar sheath, indicating a structural difference between the sheath and outer membrane. Three of the protein antigens (80, 60, and 51 kDa) were strain specific, while the other three antigens were present in other strains of *H. pylori*. Both the 51- and 48-kDa antigens were heat modifiable and likely are porins. A conserved 31-kDa protein may represent another species of porin. A method involving sucrose density ultracentrifugation and Triton extraction that allows the preparation of *H. pylori* outer membranes with minimal inner membrane contamination is described. Sodium dodecyl sulfate-polyacrylamide gel electrophoresis analysis showed that the protein content of the *H. pylori* outer membrane is similar structurally to those of other species of *Helicobacter* but markedly different from those of taxonomically related *Campylobacter* spp. and *Escherichia coli*. *H. pylori* also appeared to lack peptidoglycan-associated proteins.

Tags: Animal; Support, Non-U.S. Gov't; Support, U.S. Gov't, P.H.S.

Descriptors: \*Antigens, Bacterial--Analysis--AN; \*Bacterial Outer Membrane Proteins--Analysis--AN; \*Helicobacter pylori--Immunology--IM; Antibodies, Monoclonal--Immunology--IM; Antigens, Surface--Analysis--AN; Mice; Mice, Inbred BALB C; Molecular Weight

CAS Registry No.: 0 (Antibodies, Monoclonal); 0 (Antigens, Bacterial); 0 (Antigens, Surface); 0 (Bacterial Outer Membrane Proteins)  
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**Articles:** ☐

- ☐ Cytokine-mediated indoleamine 2,3-dioxygenase induction in response to Chlamydia infection in human macrophage cultures

AM Paguirigan, GI Byrne, S Becht, and JM Carlin

Infect. Immun. 1994. 62:1131-1136. [\[Abstract\]](#)

- ☐ Specificity and function of murine monoclonal antibodies and immunization-induced human polyclonal antibodies to lipopolysaccharide subtypes of Pseudomonas aeruginosa serogroup 06

GB Pier, NL Koles, G Meluleni, K Hatano, and M Pollack

Infect. Immun. 1994. 62:1137-1143. [\[Abstract\]](#)

- ☐ Role of polymorphonuclear neutrophil leukocytes and their integrin CD11a (LFA-1) in the pathogenesis of severe murine malaria

G Senaldi, C Vesin, R Chang, GE Grau, and PF Piguet

Infect. Immun. 1994. 62:1144-1149. [\[Abstract\]](#)

- ☐ Purification and characterization of a high-molecular-weight outer membrane protein of Moraxella (Branhamella) catarrhalis

KL Klingman and TF Murphy

Infect. Immun. 1994. 62:1150-1155. [\[Abstract\]](#)

**Identification of surface-exposed outer membrane antigens of *Helicobacter pylori*.**

**Doig P ; Trust TJ**

Department of Biochemistry and Microbiology, University of Victoria,  
British Columbia, Canada.

Infection and immunity (UNITED STATES) Oct 1994, 62 (10) p4526-33,

ISSN 0019-9567 Journal Code: G07

Contract/Grant No.: 1R01AI29927-01A2, AI, NIAID

Languages: ENGLISH

Document type: JOURNAL ARTICLE

JOURNAL ANNOUNCEMENT: 9501

Subfile: INDEX MEDICUS

Despite the potential significance of surface-localized antigens in the colonization by and disease processes of *Helicobacter pylori*, few such components have been unequivocally identified and/or characterized. To further investigate the surface of this bacterium, monoclonal antibodies (MAbs) to a sarcosine-insoluble outer membrane fraction prepared from *H. pylori* NCTC 11637 were raised. MAbs were selected on the basis of their surface reactivity to whole cells by enzyme-linked immunosorbent assay, immunofluorescence, and immunoelectron microscopy. By use of this selection protocol, 14 surface-reactive MAbs were chosen. These MAbs were used to identify six protein antigens (molecular masses, 80, 60, 51, 50, 48, and 31 kDa), all of which were localized within or associated with the outer membrane. Two of the MAbs recognized the core region of lipopolysaccharide (LPS). Only these two anti-LPS MAbs also recognized the flagellar sheath, indicating a structural difference between the sheath and outer membrane. Three of the protein antigens (80, 60, and 51 kDa) were strain specific, while the other three antigens were present in other strains of *H. pylori*. Both the 51- and 48-kDa antigens were heat modifiable and likely are **porins**. A conserved 31-kDa protein may represent another species of **porin**. A method involving sucrose density ultracentrifugation and Triton extraction that allows the preparation of *H. pylori* outer membranes with minimal inner membrane contamination is described. Sodium dodecyl sulfate-polyacrylamide gel electrophoresis analysis showed that the protein content of the *H. pylori* outer membrane is similar structurally to those of other species of *Helicobacter* but markedly different from those of taxonomically related *Campylobacter* spp. and *Escherichia coli*. *H. pylori* also appeared to lack peptidoglycan-associated proteins.

Isolation and characterization of a family of porin proteins from *Helicobacter pylori*.

Exner MM; Doig P ; Trust TJ; Hancock RE

Department of Microbiology and Immunology, University of British Columbia, Vancouver, Canada.

Infection and immunity (UNITED STATES) Apr 1995, 63 (4) p1567-72,  
ISSN 0019-9567 Journal Code: GO7

Contract/Grant No.: R01AI29927-01A2, AI, NIAID

Languages: ENGLISH

Document type: JOURNAL ARTICLE

JOURNAL ANNOUNCEMENT: 9506

Subfile: INDEX MEDICUS

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Tags: Support, Non-U.S. Gov't; Support, U.S. Gov't, P.H.S



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S3 16 S1 NOT S2

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3/9/5

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**Isolation and characterization of a family of porin proteins from *Helicobacter pylori*.**

Exner MM; Doig P; Trust TJ; Hancock RE

Department of Microbiology and Immunology, University of British Columbia, Vancouver, Canada.

Infection and immunity (UNITED STATES) Apr 1995, 63 (4) p1567-72,  
ISSN 0019-9567 Journal Code: GO7

Contract/Grant No.: R01AI29927-01A2, AI, NIAID

Languages: ENGLISH

Document type: Journal Article

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Two-dimensional gel electrophoresis was used to identify heat-modifiable outer membrane proteins, which were candidates for porins, from *Helicobacter pylori* membrane preparations. Four such proteins with apparent molecular masses of 48, 49, 50, and 67 kDa were isolated. The four proteins copurified together after selective detergent solubilizations followed by anion-exchange chromatography, and each protein was ultimately purified to homogeneity by gel purification. These proteins were then tested for pore-forming ability with a planar lipid bilayer model membrane system. All four proteins appeared to be present as monomers, and they formed pores with low single-channel conductances in 1.0 M KCl of 0.36, 0.36, 0.30, and 0.25 nS, respectively, for the 48-, 49-, 50-, and 67-kDa proteins which we propose to designate HopA, HopB, HopC, and HopD. N-terminal amino acid sequence analyses showed a high degree of homology among all four proteins, and it appears that these proteins constitute a family of related porins in *H. pylori*.

Tags: Support, Non-U.S. Gov't; Support, U.S. Gov't, P.H.S.

Descriptors: \**Helicobacter pylori*--chemistry--CH; \*Porins--isolation and purification--IP; Amino Acid Sequence; Electric Conductivity; Electrophoresis, Gel, Two-Dimensional; Heat; *Helicobacter pylori*--physiology--PH; Ion Channels--chemistry--CH; Ion Channels--isolation and purification--IP; Molecular Sequence Data; Molecular Weight; Multigene Family; Porins--chemistry--CH; Sequence Alignment; Sequence Homology, Amino Acid

CAS Registry No.: 0 (Ion Channels); 0 (Porins)

Gene Symbol: ist/GeneSymbol hopA; ist/GeneSymbol hopB; ist/GeneSymbol hopC; ist/GeneSymbol hopD

Record Date Created: 19950420

?b

**Progress in defining the inflammatory cascade.**

Figura N

School of Gastroenterology, University of Siena, Policlinico Le Scotte, Italy.

European journal of gastroenterology & hepatology (ENGLAND) Apr 1995,  
7 (4) p296-302, ISSN 0954-691X Journal Code: B9X

Languages: ENGLISH

Document type: Journal Article; Review; Review, Tutorial

Record type: Completed

Subfile: INDEX MEDICUS

*Helicobacter pylori* infection is characterized by an inflammatory response in the gastric epithelium, the intensity of which appears to be type-strain specific. Infections caused by Type I *H. pylori* organisms, i.e., those expressing VacA (the cytotoxin) and CagA (the cytotoxin-associated protein), are associated with a strong polymorph mucosal infiltration in vivo, and with increased secretion of interleukin-8 by epithelial cells. The inflammatory potential of Type II strains (non-cytotoxic, VacA- and CagA-negative) is probably less pronounced. The small urease subunit, **porins**, and other substances produced by *H. pylori* show neutrophil chemotactic activities in vitro. These bacterial components promote the adhesion of polymorphs to endothelial cells and stimulate polymorphs to generate oxygen reactive metabolites. This can severely damage the gastroduodenal mucosa. (38 Refs.)

Tags: Human

Descriptors: \**Helicobacter* Infections--metabolism--ME; \**Helicobacter pylori*; *Helicobacter pylori*--metabolism--ME; Inflammation--metabolism--ME; Interleukin-8--metabolism--ME

CAS Registry No.: 0 (Interleukin-8)

Record Date Created: 19950807

Cell surface characteristics of *Helicobacter pylori*.

Moran AP

Department of Microbiology, University College, Galway, Ireland.

FEMS immunology and medical microbiology (NETHERLANDS) Feb 1995, 10  
(3-4) p271-80, ISSN 0928-8244 Journal Code: BP1

Languages: ENGLISH

Document type: Journal Article; Review; Review, Academic

Record type: Completed

Subfile: INDEX MEDICUS

*Helicobacter pylori* is an important gastroduodenal pathogen of humans. Immunological and structural studies have been performed on the phospholipids, lipopolysaccharides (LPS) and some surface proteins of *H. pylori* strains. *H. pylori* LPS has, in general, low immunological activity and this property may aid the survival of this chronic infection. Nevertheless, *H. pylori* LPS has been found to influence the quality of gastric mucin and to stimulate pepsinogen secretion, thereby contributing to gastric disease. A number of putative adhesins of the bacterium have been described. This multiplicity of adhesins may reflect that *H. pylori* adherence is a multi-step process involving different interactions, and that different adhesins may mediate adherence to various sites in gastric tissue. (54 Refs.)

Tags: Animal; Human; Support, Non-U.S. Gov't

Descriptors: \*Bacterial Outer Membrane Proteins--chemistry--CH;  
\**Helicobacter pylori*--physiology--PH; \*Lipopolysaccharides--chemistry--CH;  
\*Lipopolysaccharides--immunology--IM; Adhesins, Bacterial--chemistry--CH;  
Adhesins, Bacterial--physiology--PH; Bacterial Capsules--physiology--PH;  
Bacterial Outer Membrane Proteins--physiology--PH; Carbohydrate Sequence;  
Cell Wall--chemistry--CH; Cell Wall--physiology--PH; Heat-Shock Proteins  
--chemistry--CH; ***Helicobacter pylori*** --pathogenicity--PY; Molecular  
Sequence Data; **Porins** --chemistry--CH; **Porins** --immunology--IM; Rats

CAS Registry No.: 0 (Adhesins, Bacterial); 0 (Bacterial Capsules); 0  
(Bacterial Outer Membrane Proteins); 0 (Heat-Shock Proteins); 0  
(Lipopolysaccharides); 0 (Porins)

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